## Remarks

The claimed invention

The present amended claims are drawn to nucleic acids comprising a modified gene including a sequence that encodes a lysostaphin protein that differs from a naturally occurring version of lysostaphin produced by a host that naturally produces lysostaphin, wherein the encoded protein includes one or more alterations with respect to the naturally occurring version of lysostaphin, and wherein one or more of the alterations disrupts one or more mammalian glycosylation events, so that the non-mammalian protein is produced and secreted by mammalian cells in its active form and is recognized by a polyclonal antibody that recognizes the naturally occurring version of the lysostaphin protein.

New independent claim 35 is drawn to a nucleic acid comprising a gene that encodes a lysostaphin protein, wherein the lysostaphin protein is recognized by a polyclonal antibody that recognizes a naturally occurring version of lysostaphin, and wherein the sequence of the lysostaphin protein contains at most one intact Asn-X-(Ser/Thr) sequences, so that the non-mammalian protein is produced and secreted by mammalian cells in its active form.

Amendments to the Claims and New Claims

Claim 1 has been amended to recite that the claimed nucleic acid comprises a modified gene including a sequence that encodes a *lysostaphin* protein that differs from naturally occurring lysostaphin such that one or more glycosylation events is disrupted, whereas the claim previously encompassed genes encoding other non-mammalian anti-staphylococcal proteins. Support for limiting the claim to lysostaphin proteins is found throughout the specification, e.g., at page 9, line 24, indicating that lysostaphin is one of the preferred proteins. In addition, the specification extensively describes the modification of naturally occurring lysostaphin and provides a reduction to practice of a modified gene encoding lysostaphin. The claim also now recites that the lysostaphin protein encoded by the modified gene is recognized by a polyclonal antibody that recognizes the naturally occurring version of the lysostaphin protein. Support for this limitation is found at page 26, line 20, through page 27, line 25, at page 31, lines 7-20, and at page 33, line 5-24, and in the figures referred to therein, in which a polyclonal anti-lysostaphin

antibody is used to detect expression of various lysostaphin proteins. In addition, one of ordinary skill in the art would recognize that binding by a polyclonal antibody is widely used as a means of determining the identity of a protein.

Claims 2 and 3 have been amended to reflect the fact that claim 1, on which they depend, claims a nucleic acid rather than a "gene". The other amendment of claim 2 is to more accurately define the invention, reflecting the fact that the gene itself comprises the mammalian regulatory element, which is operatively linked to the sequence encoding the lysostaphin protein. The other amendment to claim 3 is for grammatical purposes.

New claims 27-29 indicate that the gene can comprise a eukaryotic promoter (claim 27), which can be tissue-specific (claim 28), and/or can direct expression of the gene in mammary cells (claim 29). Support for these claims is found at page 11, line 26, through page 12, line 3. In particular, the specification lists a number of mammary-specific promoters, and Example 4 (page 34, line 10, through page 35, line 19, describes use of the β-lactoglobulin promoter to direct mammary-specific expression.

New claims 30 and 31 indicate that the gene can encode a eukaryotic secretion signal, a eukaryotic start codon, and/or the Kozak expression start site consensus sequence. Support for these claims is found at page 13, lines 1-3, page 13, lines 19-24.

New claims 32 and 33 indicate that the gene encodes a preprolysostaphin protein (claim 32) or a prolysostaphin protein (claim 33). Support is found at page 15, lines 24-28.

New claim 34, indicates that the sequence of the nucleic acid is optimized to reflect eukaryotic codon usage. Support is found at page 16, lines 17-24.

New claims 35-44 are supported by the specification and original claims in a similar manner to claims 1-3 and 27-44. New claims 35-44 avoid the use of the term "modified gene". The claims also recite the sequence of the potential mammalian glycosylation sites identified by the inventors in the naturally occurring lysostaphin sequences that they examined and require that the lysostaphin protein encoded by the claimed nucleic acids have at most one of these sites, as compared with the two sites identified in the naturally occurring lysostaphin sequences examined by the inventors.

Rejections under 35 U.S.C. § 101

Claims 1-3 stand rejected under 35 U.S.C. § 101 as being directed to non-statutory subject matter. The Examiner asserts that the claims are directed to polynucleotides which have the same characteristics as polynucleotides found naturally and therefore do not constitute patentable subject matter. While not conceding the correctness of the Examiner's position, Applicants have amended Claim 1 to include the word "isolated" as suggested by the Examiner, with the understanding that the term "isolated" shall mean that the nucleic acid (i) is removed from one or more components with which it is naturally associated and/or (ii) is modified or created by the hand of man and/or (iii) is derived either directly or indirectly from a nucleic acid having property (i) and/or (ii), e.g., by a process such as replication, transcription, amplification, etc., which process can take place either *in vivo* or *in vitro*.

The instant application describes isolated nucleic acids having the properties recited in claim 1, as is evident from the Examples and the specification as a whole, thus providing support for the Amendment. As described in Examples 1 and 3, the increased purity of the claimed nucleic acids relative to the form in which nucleic acids are found in nature allows them to be manipulated *in vitro* and to be inserted into cloning vectors. The increased purity of the claimed nucleic acids relative to the form in which nucleic acids are found in nature also facilitates their use in the construction of transgenic animals, as described in Examples 4 and 5.

Rejections under 35 U.S.C. § 112

Claims 1-3 stand rejected under 35 U.S.C. § 112, first paragraph, on the ground that the specification lacks sufficient written description. The Examiner states that "the disclosure fails to describe the common attributes or characteristics that identify the members of the genus" that constitutes the claimed invention and that because the genus is highly variant, the term "modified gene" is insufficient to describe the genus. The Examiner asserts that the specification does not "clearly allow persons of ordinary skill in the art to recognize that [he or she] invented what is claimed." While not conceding the correctness of the Examiner's position, Applicants respectfully assert that the rejection does not apply to the claims as amended, for each of the following reasons.

Firstly, the disclosure does describe the common attributes and characteristics that identify members of the genus of modified genes as presently claimed. These features, which

are further described below, include the following: (i) The modified genes include a sequence that codes for a lysostaphin protein; (ii) The lysostaphin protein differs from a naturally occurring version of lysostaphin produced by a host that naturally produces the lysostaphin protein. In particular, the encoded sequence includes one or more alterations with respect to naturally occurring lysostaphin, resulting in disruption of one or more mammalian glycosylation events; (iii) The non-mammalian protein is produced and secreted by mammalian cells in its active form. The claim is limited to nucleic acids that encode proteins that are recognized by a polyclonal antibody that recognizes naturally occurring lysostaphin, thus restricting the number of possible variants and providing the structural limitation that the Examiner asserts was lacking in the previous version of the claim. Only nucleic acids that encode proteins having a sequence sufficiently similar to that of naturally occurring lysostaphin to result in recognition by a polyclonal antibody that recognizes naturally occurring lysostaphin fall within the claimed genus.

Each of the lysostaphin proteins encoded by the claimed nucleic acids contains an alteration that disrupts a mammalian glycosylation event. As described throughout the specification, e.g., at page 14, line 11, through page 15, line 10, Applicants discovered that glycosylation prevents production of an active, secreted lystostaphin protein by mammalian cells, as occurs with bacterial hosts. Applicants further discovered that preventing such glycosylation (e.g., by altering one or more potential mammalian glycosylation sites), allows for the production of active lysostaphin by mammalian cells. One of ordinary skill in the art would recognize that glycosylation events in mammalian cells occur at sites having the sequence Asn-X-(Ser/Thr), as described in the specification at page 14, line 19. These sites occur at well-defined positions within the lysostaphin sequence, which correspond to well-defined positions within the claimed nucleic acids that encode the lysostaphin proteins. In accordance with the invention, an alteration (e.g., substitution, deletion, or addition) in the amino acid sequence disrupts a glycosylation event that would otherwise occur within mammalian cells. The known location of these sites, and the teaching that the claimed nucleic acids encode a lysostaphin protein in which a glycosylation event at one or more of these sites is disrupted, provide a further structural limitation.

The specification describes two potential mammalian glycosylation sites having the sequence Asn-X-(Ser/Thr) in lysostaphin. The specification teaches the replacement of one or

both Asn residues with a different amino acid, of which only 20 exist in mammalian cells (page 14, line 23, through page 15, line 6). The specification provides a specific nucleic acid sequence (SEQ ID NO: 3) that encodes a lysostaphin protein in which both Asn residues are replaced by Gln, and describes the alteration of AAT codons to CAG codons in order to replace Asn by Gln in the encoded protein (Example 1, at page 24, line 10, through page 25, line 19). The disclosed species is representative of the genus in that it disrupts a specific glycosylation event. The specification further teaches that additions or deletions would also alter the site. Thus the specification implicitly discloses numerous additional nucleic acid sequences that meet the limitations of the claims. Whether or not any particular sequence results in a lysostaphin protein that is secreted by mammalian cells in an active form can readily be determined using the assays described in Examples 1 and 3 and others known in the art. Thus Applicants submit that the specification would clearly allow one of ordinary skill in the art to recognize that Applicants were in possession of the claimed invention.

Secondly, Applicants submit that the instant claims differ from those at issue in *Lilly* in an important respect that makes a direct application of *Lilly* inappropriate in this case. In *Lilly*, the claims involved a genus or species of nucleic acids, namely cDNAs having the structure of a reverse transcript of *vertebrate* mRNA, *mammalian* mRNA, or *human* mRNA, based purely on the functional limitation that the mRNA encode insulin and the disclosure of the rat cDNA sequence. Unlike the instant claims, the exact nucleotide sequences were a key feature of the claims, since only specific, predetermined sequences (i.e., vertebrate, mammalian, or human) would meet the claim limitations. Knowledge of the rat sequence could not provide a description of additional members of the vertebrate genus or the mammalian genus and could not provide a description of the human species. Until the claimed vertebrate, mammalian, or human mRNA sequences had actually been determined, there was no way to determine whether any particular nucleic acid sequence fell within the scope of the claims. In other words, a large number of sequences might readily have met the purely functional limitation, i.e., encoding insulin, while failing to correspond identically with the actual vertebrate, mammalian, or human nucleic acids that were claimed. Thus the functional limitation was insufficient to describe either the genus or the species and it was arguably reasonable to require disclosure of a representative number of specific nucleic acid sequences to support the generic claims and to require disclosure of the exact human cDNA sequence to support the species claims. In contrast, in the instant application

the exact nucleotide sequences of nucleic acids yet to be isolated are not required, provided that the various structural and functional limitations described above are met. Rather, one of ordinary skill in the art can readily determine numerous specific nucleic acid sequences that meet the limitations of the claims by modifying the known nucleic acid sequences of naturally occurring lysostaphin at specific, known locations, resulting in nucleic acids with known sequences, as clearly described in the specification.

Thirdly, notwithstanding the fact that *Lilly* is not directly applicable to the instant claims, Applicants submit that the requirements set forth in the Revised Interim Guidelines for the Examination of Patent Applications Under 35 U.S.C. 112, 1 "Written Description" Requirement (hereinafter "Written Description Guidelines") referred to by the Examiner are fully met by the instant application, as discussed below.

Applicants have disclosed that species falling within the genus are recognized by a polyclonal antibody that recognizes naturally occurring lysostaphin, thus providing a structural limitation that correlates with the functional requirement that the claimed nucleic acid encodes a lysostaphin protein and represents a physical/chemical property of the claimed nucleic acids. Polyclonal antibodies that recognize naturally occurring lysostaphin are well known in the art (see, e.g., Gagne, cited by the Examiner, Williamson, cited previously by the Examiner), and the specification describes use a representative antibody to detect lysostaphin using a variety of different immunologic methods at page 26, lines 20-27, at page 31, lines 7-20, and at page 33, line 5-24.

Applicants have further disclosed that all species falling within the genus contain an alteration that disrupts one or more mammalian glycosylation events and have specifically disclosed the potential mammalian glycosylation sites in lysostaphin, thus providing a structural limitation that correlates with the functional limitation that the lysostaphin protein is secreted by mammalian cells in active form. Applicants have provided extensive guidance as to various methods by which such sites may be modified in order to disrupt glycosylation, e.g., by making a substitution at the Asn residue that is actually glycosylated, or by making a deletion or addition that disrupts the Asn-X-Ser/Thr sequence. Unlike the case presented in Example 13 of the Written Description Guidelines, in which a claim to a "protein variant" lacked adequate written description where "the specification and claim do not provide any guidance as to what changes should be made", the instant specification and claims do provide such guidance. The

specification and claims further provide structural limitations as described above and distinguishing features and common attributes of the claimed invention. As in Example 15 of the Written Description Guidelines, in which the written description requirement was met, given the teachings of the instant specification, "one skilled in the art would be able to immediately envisage members of the genus embraced by the claim."

Applicants have described various elements that optionally may be included in the modified genes of the instant claims, e.g., eukaryotic signal sequences, eukaryotic promoters, eukaryotic translation initiation sequences such as the Kozak consensus sequence, polyadenylation signals, etc. See, e.g., page 11, line 14, through page 14, line 2. Applicants further submit that these terms are well known in the art and that one of ordinary skill in the art, reading the terms themselves, would readily understand their meaning without additional written description. As the Federal Circuit has stated, "A patent need not teach, and preferably omits what is well known in the art." *Spectra Physics, Inc. v. Coherent, Inc.*, 827 F.2d 1524, 3 USPQ2d 1737 (Fed. Cir. 1987).

Furthermore, Applicants have described the complete structure of a representative species falling within the genus of modified genes of claim 1 (SEQ ID NO:3) and have described a method of making the gene (see Example 1). As stated in the Written Description Guidelines at page 31, "The disclosure of a single disclosed species may provide an adequate written description of a genus when the species disclosed is representative of the genus." Here, the features common to the members of the genus constitute a substantial portion of the genus. Unlike Example 7 of the Written Description Guidelines, in which disclosure of an EST sequence did not adequately describe a genus of cDNAs of which the EST was only a (possibly non-coding) fragment, the representative species explicitly disclosed by Applicant embodies significant structural limitations and attributes shared by members of the genus, and these structural limitations and attributes outweigh any variability within the genus. Applicants submit that the disclosed species is representative of the genus and point out that the Examiner has not pointed to any evidence to suggest that this is not the case. Furthermore, as pointed out above, the specification implicitly discloses numerous other members of the genus by describing that one or more of the glycosylation site(s) can be altered by replacement with amino acids other than Gln, e.g., by making conservative substitutions. The Written Description Guidelines

recognize that a sufficient number of species to support a generic claim can be implicitly or explicitly disclosed (see Written Description Guidelines, p. 9).

Reduction to practice of the implicitly disclosed species and testing them to determine whether they meet the functional limitations of the claims is straightforward, given the teachings of the specification (e.g., assays for lysostaphin activity, described at page 26, line 7-15 and elsewhere in the specification) and the knowledge of one of ordinary skill in the art. Representative methods of making numerous members of the claimed nucleic acid genus are clearly described, although it is to be understood that other methods of altering the sequence so as to prevent glycosylation are also encompassed. For example, starting with a sequence that encodes naturally occurring lysostaphin, the artisan designs a sequence that encodes a protein in which an amino acid residue located at a mammalian glycosylation site is either eliminated or substituted, or in which amino acid addition to disrupt the site occurs. The specification provides a nucleic acid sequence that encodes naturally occurring lysostaphin (SEQ ID NO: 1). The specification further provides amino acid sequences for several naturally occurring lysostaphin proteins (SEQ ID NOs: 2, 5, and 6). One of ordinary skill in the art can readily derive nucleic acid sequences that encode the proteins of SEQ ID NOs 2, 5, and 6, to use as starting materials or can use SEQ ID NO: 1. The nucleic acids containing modified genes can be made by methods that are conventional in the art, including complete chemical synthesis or PCR (see Example 1). It is noted that additional methods can also be used, e.g., mutagenizing host cells that express naturally occurring lysostaphin and screening for variants.

In summary, the specification describes at least two structural limitations on the claimed invention, i.e., the requirement that the claimed nucleic acids encode lysostaphin proteins that are recognized by polyclonal antibodies that recognize naturally occurring lysostaphin, and the requirement that the claimed nucleic acids encode lysostaphin proteins in which a glycosylation event at a specific site (i.e., Asn-X-Ser/Thr) is disrupted. These structural limitations correlate with functional characteristics shared by members of the genus, i.e., the requirements that the nucleic acid (i) encodes a lysostaphin protein and (ii) that the protein is secreted by mammalian cells in its active form. Furthermore, a method of making the claimed nucleic acids and appropriate starting materials are clearly described. Applicants have thus provided partial or complete structure, physical or chemical properties (recognition by an antibody), functional characteristics and a correlation between structure and function, and a method of making, i.e., all

the distinguishing identifying characteristics set forth in the Written Description Guidelines (see p. 8). Applicants thus submit that the written description requirement is met with respect to the amended claims and respectfully request withdrawal of the rejection.

Rejections under 35 U.S.C. § 102

Claim 1 stands rejected under 35 U.S.C. § 102(b) as being anticipated by Gagne, WO 96/37793 (hereinafter "Gagne"). The Examiner takes the position that a gene created by altering a gene encoding a protein with anti-staphylococcal properties such as β-lytic protease, e.g., by deleting all amino acids (*sic*) and substituting with amino acids corresponding to lysostaphin, thereby resulting in a nucleic acid comprising a "modified gene", falls within the scope of the claimed invention and is therefore anticipated by Gagne. While Applicants respectfully disagree with this rejection and reserve the right to traverse it in a future application, it is submitted that the amendments to the claims render the rejection moot.

The instant claims are drawn to a nucleic acid comprising a modified gene including a sequence that codes for a lysostaphin protein *that differs from a naturally occurring version of lysostaphin* produced by a host that naturally produces lysostaphin. The encoded protein includes one or more alterations with respect to naturally occurring lysostaphin, thus requiring that the nucleic acid encoding the protein includes one or more alterations with respect to a nucleic acid that encodes naturally occurring lysostaphin. The one or more alterations disrupt one or more mammalian glycosylation events. At most, Gagne discusses nucleic acids that encode only the naturally occurring form of lysostaphin produced by *Staphylococcus simulans*. See, for example, page 39, lines 26-29, describing that the lysostaphin gene was obtained by PCR amplification from extracted DNA of *Staphylococcus simulans* biovar *staphyloliticus*. Gagne nowhere teaches or suggests modification of the natural lysostaphin-encoding sequence he employs. Thus Applicants submit that Gagne cannot anticipate the instant claims, which explicitly require such modification. Withdrawal of the rejection is respectfully requested.

In conclusion, in view of the amendments and remarks presented herein, none of the cited art anticipates any of the claims pending in the instant application, and the application complies with the requirements of 35 U.S.C. §§ 101, 102, and 112. Applicants therefore respectfully submit that the present case is in condition for allowance. A Notice to that effect is respectfully requested.

If, at any time, it appears that a phone discussion would be helpful, the undersigned would greatly appreciate the opportunity to discuss such issues at the Examiner's convenience. The undersigned can be contacted at (617) 248-5000 or (617) 248-5175 (direct dial).

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Respectfully submitted,

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